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A key barrier to our understanding of the organizational principles underlying neuronal and neural circuit computations has been the impenetrable complexity of the cortical network. To begin addressing this complexity we examined the interplay between circuit structure and neuronal function of the sparsely connected pyramidal cell network of the visual cortex. We use volumetric *in vivo* two-photon calcium imaging of a genetically-encoded calcium indicators to measure sensory physiology of a large population of identified neurons and subsequently reconstructed the local excitatory neuronal network using large-scale EM. Our recent results suggest that wiring specificity acts as a substrate for computations underlying cortical sensory processing.

By combining *in vivo* physiology in awake, behaving animals and large-scale electron microscopy (EM), we are uniquely poised to examine functional connectomics in neocortical circuits.

While recent advances in large-scale EM have allowed us to begin relating circuit structure to neuronal function, even at our current fast rates it would take ~1 year to manually prepare and ~10 years to image a complete local cortical circuit – a mammalian cortical column – at the resolution required to unambiguously extract network connectivity. Clearly, the speed of data acquisition continues to be a critical barrier. We are addressing this by developing a pipeline utilizing a novel substrate for automated sample collection compatible with high-speed transmission EM (TEM) imaging. We refer to this substrate as *Grid-Tape*. Grid-Tape will allow us to leverage advances in both reliable, automated sample collection and high-quality, large-scale image acquisition.

With this new methodology and instrumentation, we expect to increase the speed of EM data acquisition by up to two orders of magnitude compared to conventional TEM systems. This would allow high-quality nanometer-resolution imaging of a local cortical circuit in ~1 year. We will immediately apply this approach to increase our understanding of the fundamental principles underlying cortical processing and organization. Moreover, we will have the ability to examine neuronal connectivity across multiple individuals to define the precision and stereotypy of connections, giving insight into developmental and experience-dependent mechanisms guiding functional circuit formation and plasticity

We are interested in the generation and analysis of integrated functional and EM-connectivity data to discover principles underlying computations and information processing in neuronal networks.

Fig. 1. Visual Cortical Function & Network Anatomy

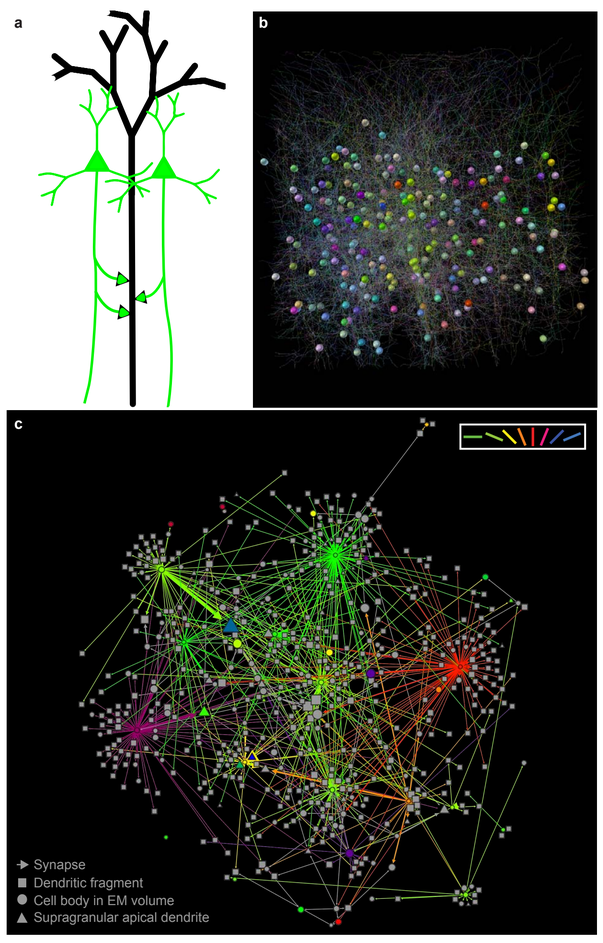
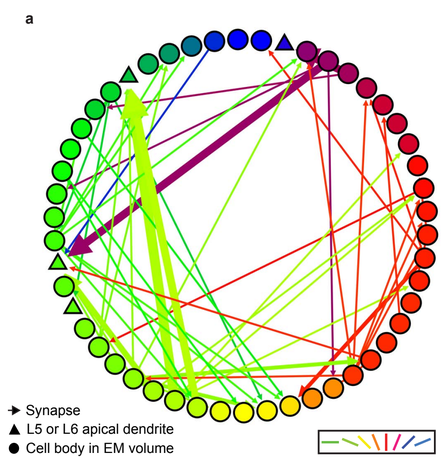


Fig. 2. Visual Physiology Predicts Exictatory Connectivity



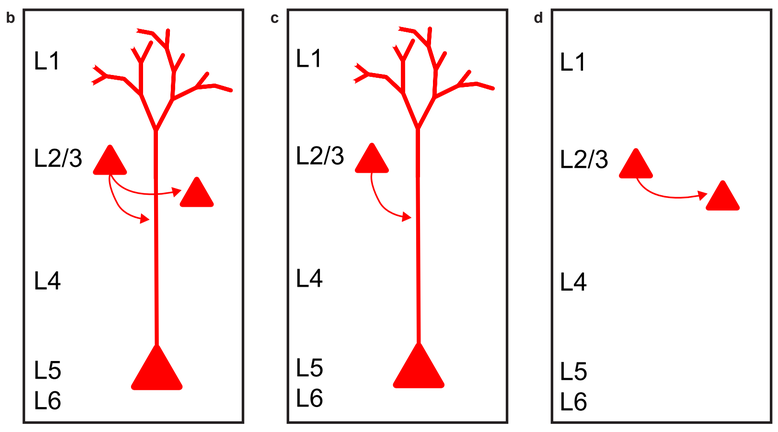


Fig. 3. Receptive Field Properties & Convergent Connectivity

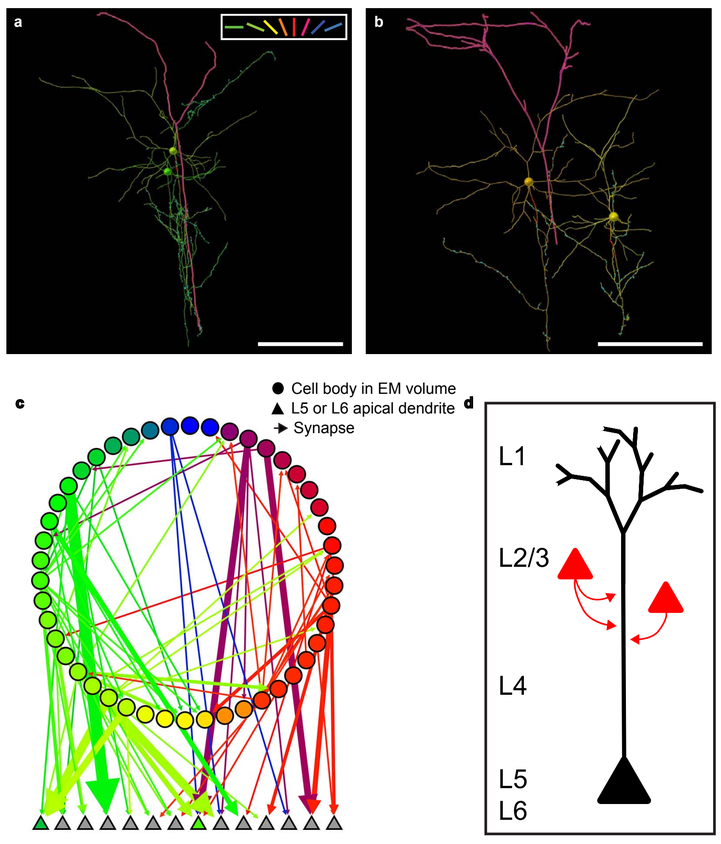


Fig. 4. Grid-Tape: A Substrate for Automating High-Throughput Transmission Electron Microscopy

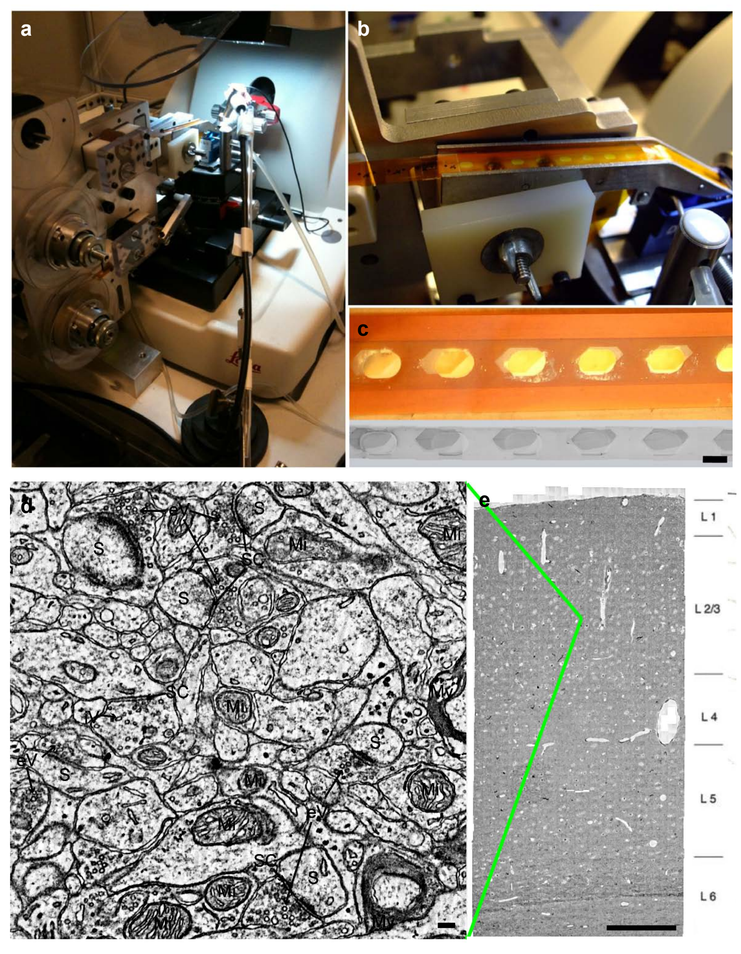
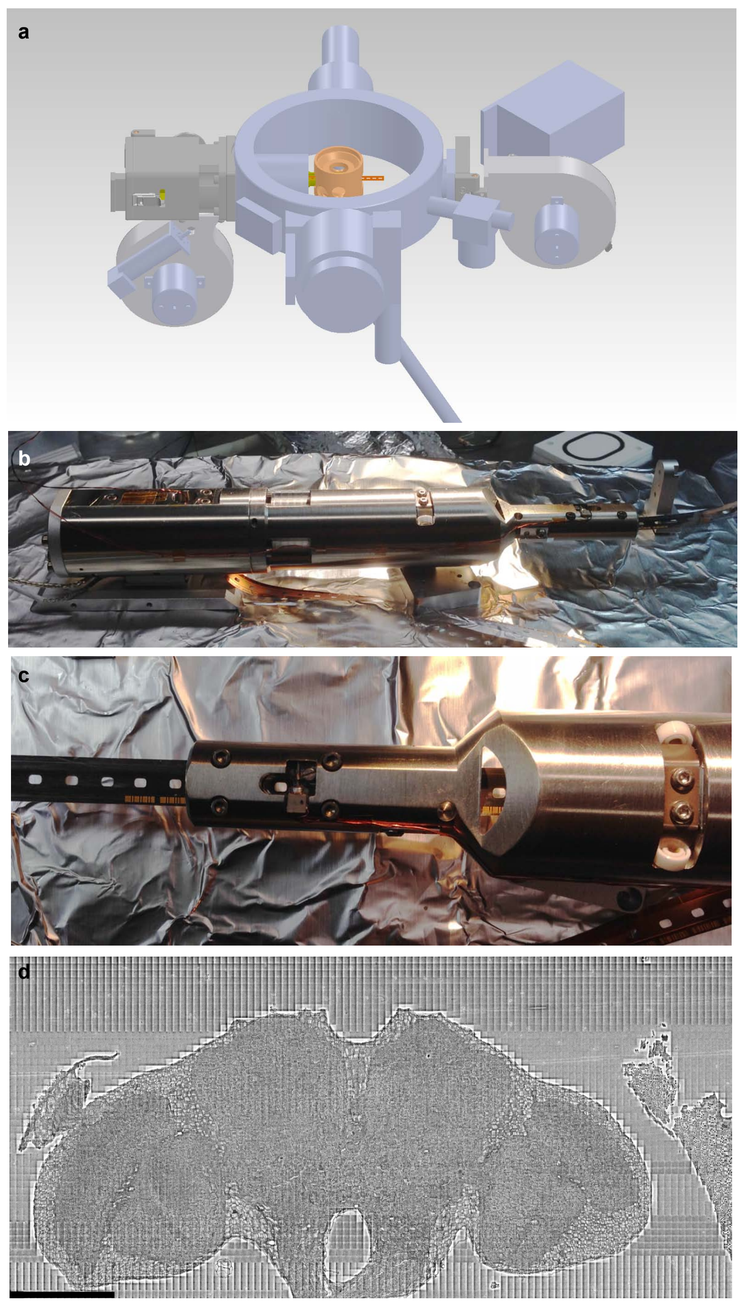


Fig. 5. TEMCA-GT: Automating Large-Scale Transmission Electron Microscopy Imaging



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